# Use of Data to Inform Risk Characterization and Management in Addressing Biofilm Problems

Dr Nick Ashbolt NERL, U.S. EPA, Cincinnati January 31, 2007

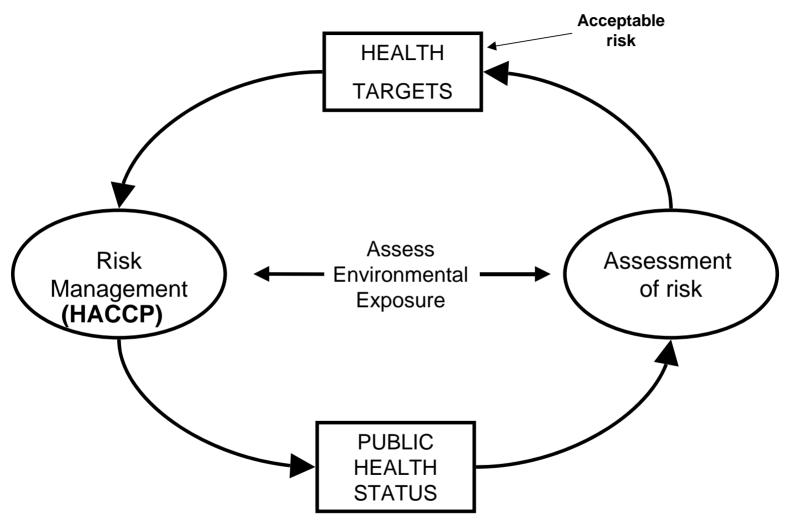
# WHO risk-based approach to guidelines

- Fewtrell & Bartram (2001)
   Guidelines, Standards and Health
  - www.who.int/water\_sanitation\_h ealth/dwq/whoiwa/en/
- WHO 3<sup>rd</sup> Edition of Drinking Water Guidelines (2004)
  - http://www.who.int/water\_sanitati on\_health/dwq/en/index.html





#### WHO Risk management approach

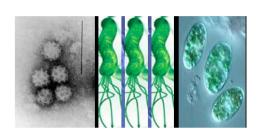


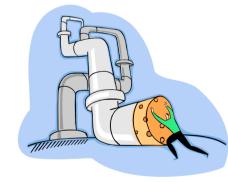
Fewtrell & Bartram (2003) IWA Publishing



### Risk characterisation associated with distribution

- Requires information on:
  - Hazards (pathogens: fecal + non)
  - Hazardous events (breaks, backflow, X-connection, sloughing, etc.)
  - Hazard doses (conc. X vol.)
  - Dose-response (probability of illness)









# Risk estimates (inf. person<sup>-1</sup>.y<sup>-1</sup>) for simulated distribution intrusion event (MicroRisk Project)

Simulation	Avg.	95%ile	99%ile
Baseline (Campylobacter spp.)	4.9 10-8	1.8 10-7	1.310-6
Baseline + Event (Acute) Infiltration of <i>E. coli</i> PDF T(0.5,1,10); Ratio of <i>E. coli : Campylobacter</i> = 1000; Probability of being affected = 0.00031 person <sup>-1</sup> .d <sup>-1</sup> .	1.8 10-5	6.5 10-5	4.8 10-4
Duration of Event = 3 days			

Number in bold above Dutch benchmark risk of 10<sup>-4</sup>.person<sup>-1</sup>.year<sup>-1</sup>



#### Points covered



- 1. Criteria for identifying a significant biofilm problem
- Concerns that make biofilms a problem
   & some aspects for our advantage
- 3. Possible strategies for mitigating the biofilm problem



# 1. Criteria for identifying a significant biofilm problem

Traditional - increase in:



- HPCs
- TCs
- dirty water
- taste & odor
- loss of chlorine residual
  - Other use of SCADA data

Each significant if lack of compliance or complaints



# Growth of coliforms in biofilms – may not be a health issue

- Various coliforms are known to grow in pipe biofilms
  - Incl. Citrobacter, Enterbacter, Klebsiella
  - Latter include fecal coliform members
  - Hence importance of using E. coli or enterococci as the fecal indicator in follow-ups
- Some 3-8 % of systems have MCL non-acute violations but only 10% of these have acute violations (*E. coli* presence)

But how to separate growth from an intrusion event ...

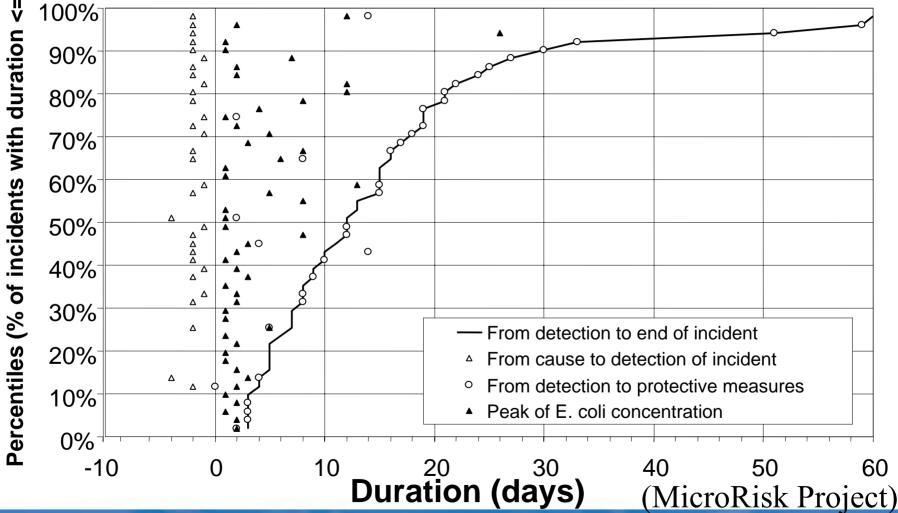


#### Fecal intrusion event: Investigation of taste issues 3<sup>rd</sup> Dec

Date	Homes	No of samples containing indicator bacteria &				
(2001)	Sampled	sampled (max concentration CFU/100mL)				
	(100mL)	Coli37	Coli44	SSRC	FS	
Dec. 4	2	<b>2</b> of 2 (19)	-	-	-	
Dec. 5	12	<b>9</b> of 12 (13)	<b>8</b> of 12 (5)	<b>1</b> of 2 (4)	<b>2</b> of 12 (4)	
Dec. 6	28	<b>8</b> of 28 (14)	<b>5</b> of 27 (9)	<b>8</b> of 22 (5)	<b>2</b> of 22 (3)	
Dec. 7	19	0 of 19	0 of 19	0 of 19	0 of 19	
Dec. 8	7	0 of 7	0 of 7	0 of 7	0 of 7	
Dec. 9	21	0 of 21	0 of 21	<b>1</b> of 21 (1)	0 of 21	
Dec. 10	23	0 of 23	0 of 23	0 of 21	0 of 23	
Dec. 11	5	0 of 5	0 of 5	0 of 5	0 of 5	
Dec. 12	12	0 of 12	0 of 12	<b>1</b> of 12 (1)	0 of 12	
Dec. 13	6	0 of 6	0 of 6	0 of 6	0 of 6	
Dec. 14	10	0 of 10	0 of 10	0 of 10	0 of 10	
Dec. 15	13	0 of 13	0 of 13	0 of 13	0 of 13	
Total	158	17 of 156	13 of 155	11 of 138	4 of 150	



# Duration of 50 fecal contamination incidents (Netherlands: 1994-2003 by 7 companies supplying c. 11 million affecting c. 185,000 inhabitants)





### Interpreting persistent non-acute TCR MCL violations

- In the absence of E. coli
  - Generally considered a non-health issue (from fecal pathogens)
  - However, if moderate chlorine residuals, fecal pathogens could still be infectious
     & E. coli non-culturable
  - Quick PCR test for E. coli, Clostridium perfringens (spores) or other persistent fecal marker could resolve this dilemma



# High TC & no E. coli – what about opportunistic pathogens?

- This is the real question for which there is no strong evidence of health effects, but no serious study either!
- No science to suggest that TC are a good index of opportunistic pathogens
- Rather just one of a group of heterotrophic bacteria that may grow in biofilms



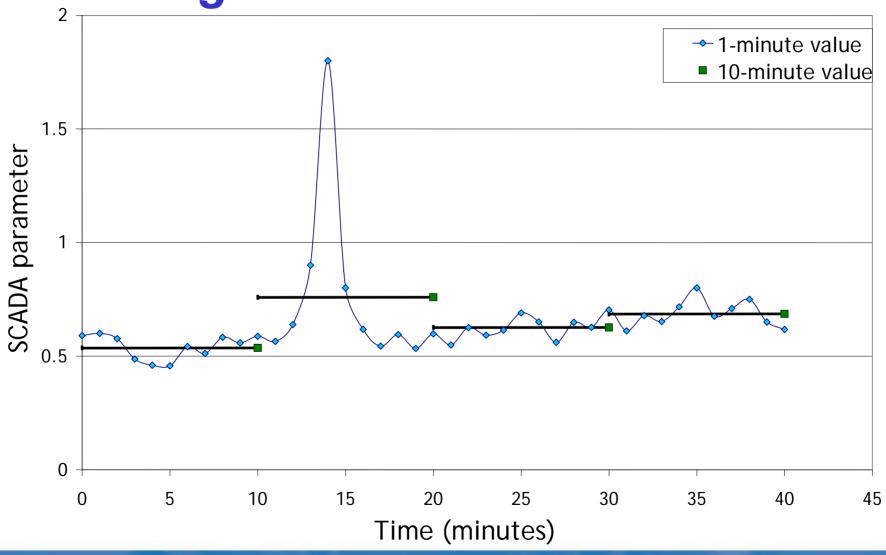
# Emerging criteria to ID biofilm problems (control charting at CCPs)

- On-line measurements that trend over critical limits, e.g.
  - Chlorine residual loss
  - Nitrification (nitrite)
  - ATP, TOC or other biomass measures
- Biofilm coupon assessment
  - In-pipe, annular reactors or loops off-pipe
    - ATP, TOC or other biomass measures
    - Rapid community 'fingerprinting'





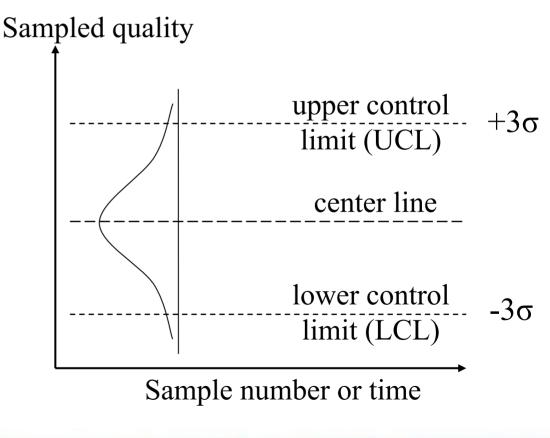
# Identifying a hazardous event: e.g. 1 & 10 min SCADA data





# Basis of control charting & Change point analysis

- Change point analysis used to detect slight changes missed by control charts
  - Cumulative Sum Control Charting (CUSUM)





#### **CUSUM** charts

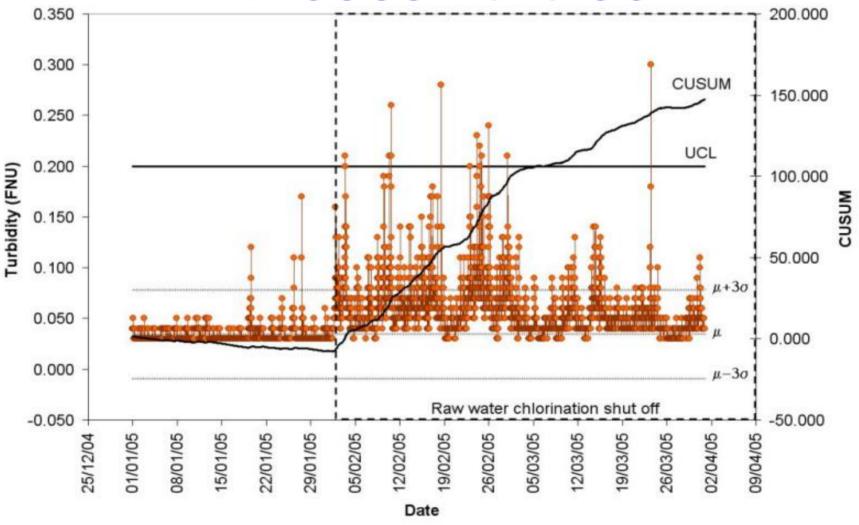
 The simplest CUSUM charts are constructed from a cumulative sum (S<sub>n</sub>) based on the data:

$$S_n = S_{n-1} + (X_n - \overline{X})$$

- n is the total number of data points,  $X_n$  the data point, and  $\overline{X}$  the arithmetic mean of the data points
- A CUSUM chart is interpreted as follows:
  - An upward slope indicates a period with values above average
  - A downward slope indicated a period with values below average
  - A sudden change in direction indicates a shift or change in the average
  - If the chart follows a straight path this indicates a period where the average did not change



### Filtrate water turbidity SCADA with CUSUM and UCL



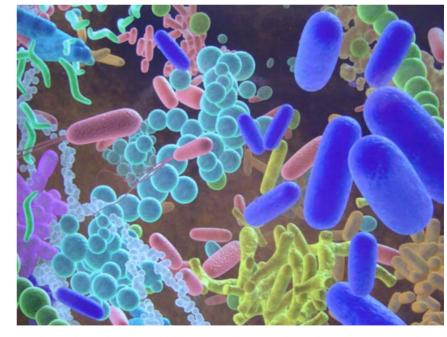


# 2. Concerns that make biofilms a problem

 Biofilms sequester fecal pathogens and allow the growth of opportunistic

pathogens

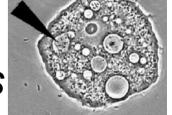






### Water-based bacterial pathogens

- Various Legionella strains
- Mycobacterium avium, M. ulcerans



- Burkholderia pseudomallei
- Helicobacter pylori
- Aeromonas & Vibrio spp.
- Campylobacter spp.?
- All grow associated with amoeba in biofilms & may be active but non-culturable



# Pathogens also protected in biofilm ecosystems

- Biofilm slime 'mops-up' chlorine disinfectants & pathogens
- Acanthamoebae cysts remained viable
  - after treatment with 100 mg/L chlorine (free and combined) for 10 min, as well as
  - 80°C for 10 min containing viable legionellae
- Implying that conventional hyper-disinfection or 80°C heating may be insufficient for longterm control of Acanthamoebae-bound Legionellae in water distribution systems

Storey et al. (2005) Scand. J. Infect. Dis. 36(9):656-662



#### And it gets worse!

- Acanthamoeba polyphaga Mimivirus largest known DNA virus
- The word "girus" used to recognize the intermediate status of these giant **DNA** viruses
- 750 nm dia
- genome complexity which is closer to small parasitic prokaryotes than to regular viruses<sup>1</sup>

 Possibly > legionellae in causing community & nosocomal pneumonia<sup>2</sup>

> <sup>1</sup>Claverie et al. (2006) Virus Res.117(1):133-44 <sup>2</sup>La Scola *et al.* (2005) Emerg. Inf. Dis. 11(3):499-52 <sup>2</sup>Berger *et al.* (2006) Emerg. Inf. Dis. 12(2):248-55



### Unintended disinfection effects

- Disinfection stresses cells, producing viable but non-culturable bacteria on selective media ( = false negatives)
  - Yet would be PCR-positive (false positives)
- Hence, a molecular method or a chemical / chlorine-resistant fecal microbe could be used to indicate a contamination/biofilm event
  - e.g. on-line PCR, loss of chlorine residual, fecal sterol, change in NH<sub>3</sub> or clostridial spore assay



## Whereas biofilms provide a history of contamination

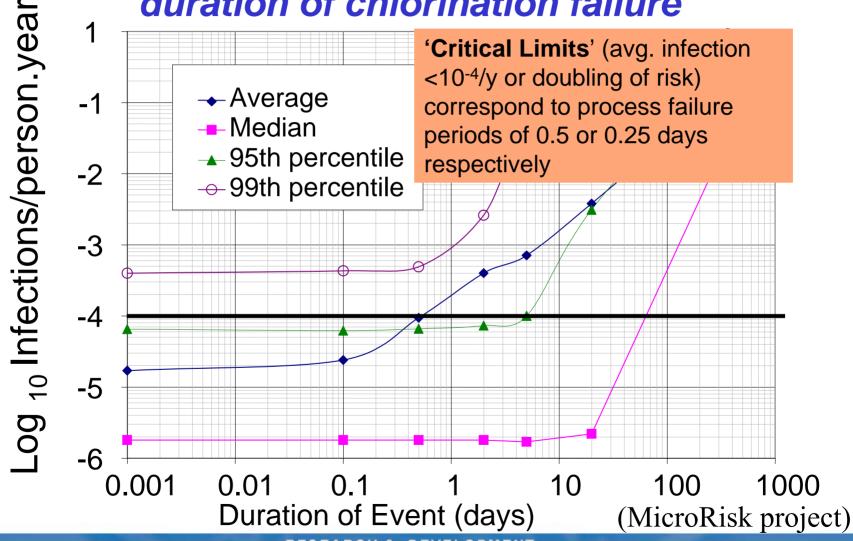
- Because of their sequestering nature, biofilms are a good integrator of passed contamination
- Hence, biofilms may provide a
   preferable target to monitor than water

   more representative, particularly for
  - small systems with infrequent sampling



#### Short-term events are important

Campylobacter annualised infection risk vs duration of chlorination failure





# 3. Possible strategies for mitigating the biofilm problem

- Once established, biofilms can readily reimmerge as a problem
  - As shown for Legionella in buildings and nitrification in chloraminated systems
- Control of biofilms means control of the factors for biofilm growth:
  - Temperature, C, N, P
  - But even very low C, N or P can yield problems in warm waters
  - Disinfection ± (e.g. monochloramine for Legionella, but may increase Mycobacteria)
  - Selection of beneficial biofilm members requires community/ecological studies



# Hence, do not let biofilms establish in the first place

- Regular mains cleaning preventative
  - Reduce dead ends and stagnant water zones
- But if biofilms reach the 'pain threshold':
  - Change disinfectant (however » dirty water)
  - Mains flushing/pigging
  - Do not replace with iron piping



# International best management practice

- WHO/NHMRC: Neither HPC nor TC's promoted as fecal indicators, but may indicate possible biofilm problems (primarily indicators of water treatment disinfection efficacy)
- Water safety plans used: in distribution meaning focus on-line for changes in chlorine residuals, NH<sub>3</sub>, TDS, turbidity or pressure



#### What is needed

- Inclusion of biofilm assessment as a key component for distribution system management, but reliant on:
  - New knowledge on ecology of fecal and non-fecal pathogens in pipe biofilms
    - Health significance still a major data gap
  - Sanitary survey and on-line control of hazardous events more important than sampling and culture-based monitoring



### Acknowledgments & Disclaimer

- I would like to acknowledge the input of many UNSW students and colleagues who have worked with me on distribution systems & HACCP
- This presentation does not necessarily reflect official Agency policy

